Development and Evaluation of Innovative Tools:

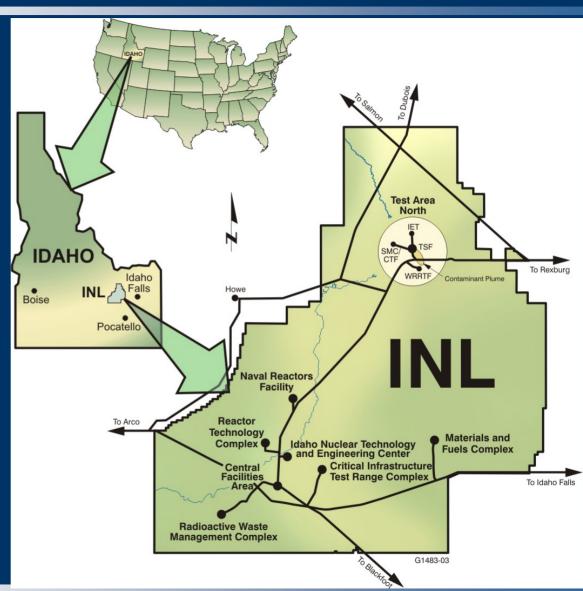
Improvements to Long-Term Performance and Cost at the Test Area North Groundwater Plume

M. Hope Lee



Historical Background

- Industrial
 wastewater was
 directly injected into
 the aquifer from
 1953-1972.
- Primary contaminant of concern is TCE.
- TCE plume is nearly 2 miles long.
- Contaminated aquifer is 200-400 ft deep.
- Aquifer is comprised of fractured basalt.





Regulatory Background

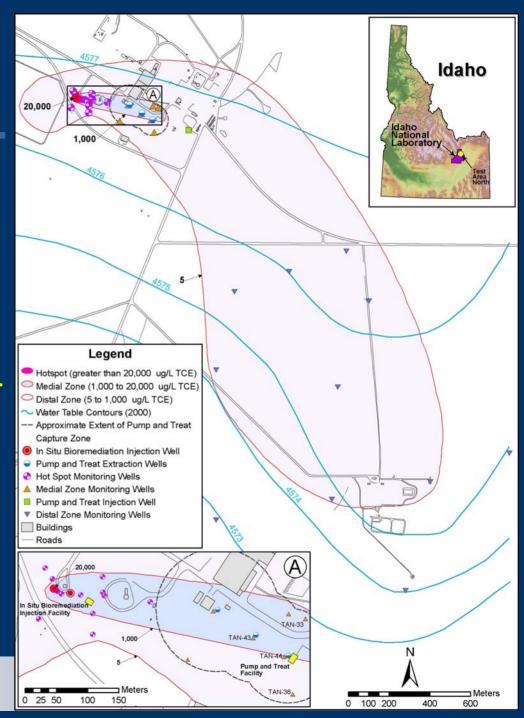
- 1995 Record of Decision
 - Selected pump and treat (P&T) as the default remedy, but allowed for alternative technology evaluations
 - Established 100 year restoration timeframe (2095)
- 1997 Explanation of Significant Differences
 - Defined three plume zones
 - Performed alternative technology evaluations
- 2001 Record of Decision Amendment
 - Identified alternative remedies for two of the three plume zones



Three-Component Remediation Strategy

- Hotspot
 TCE >20,000 μg/L
 In Situ Bioremediation
- Medial Zone
 TCE 1,000-20,000 μg/L
 Pump and Treat
- Distal Zone
 TCE <1,000 μg/L
 Monitored Natural
 Attenuation





Outline for Discussion

- Hotspot
 Improvements in In Situ Bioremediation Strategy
 - Enhanced Dissolution
 - Area of Influence
- Medial Zone
 Evaluation of alternative remediation technologies
 - Pump and Treat Performance
 - Biological Attenuation
 - In situ evaluations of biological degradation



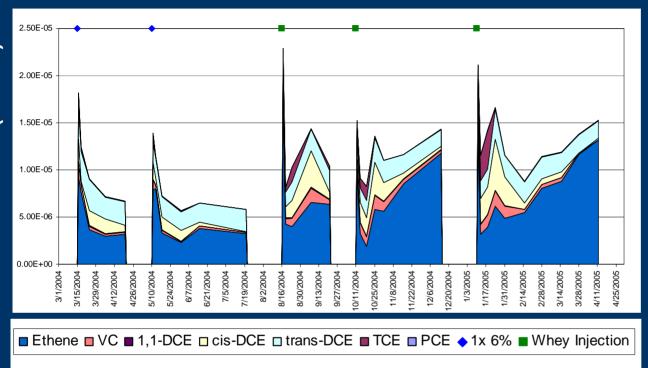
Hotspot: Evaluation of Electron Donors

Purpose: To improve performance and decrease cost

- 1. Laboratory studies of 6 electron donors
- 2. Field comparison of sodium lactate vs. whey powder
- Anaerobic degradation results in rapid degradation of aqueous phase contaminants.
- Dissolution of the residual source material to the aqueous phase is the rate-limiting factor for cleanup at TAN ...
- To optimize clean-up, need to maximize dissolution of source material while stimulating effective biodegradation of liberated contaminants.



Enhanced Dissolution of Residual Source

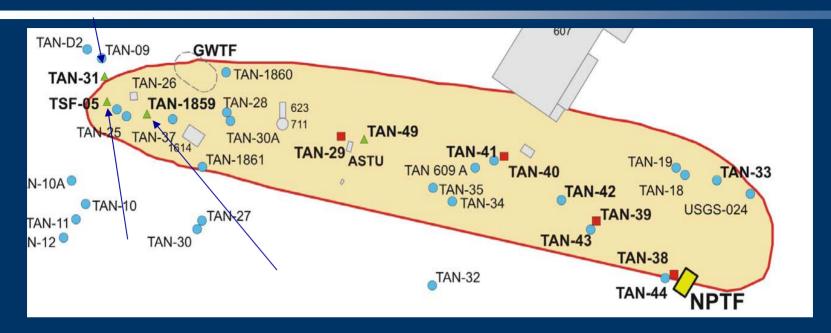


- Electron donor distribution, utilization, & cost
- Dechlorination and dissolution of TCE from residual source
- Molecular characterization of communities in situ

Increased enhanced dissolution of the source following whey powder injections, as compared to sodium lactate injections.



Increase Area of Influence



- Multiple well injections
- Outfitted new wells on the periphery of the hotspot for injection capability
- Monitor distribution of electron donor and associated source material

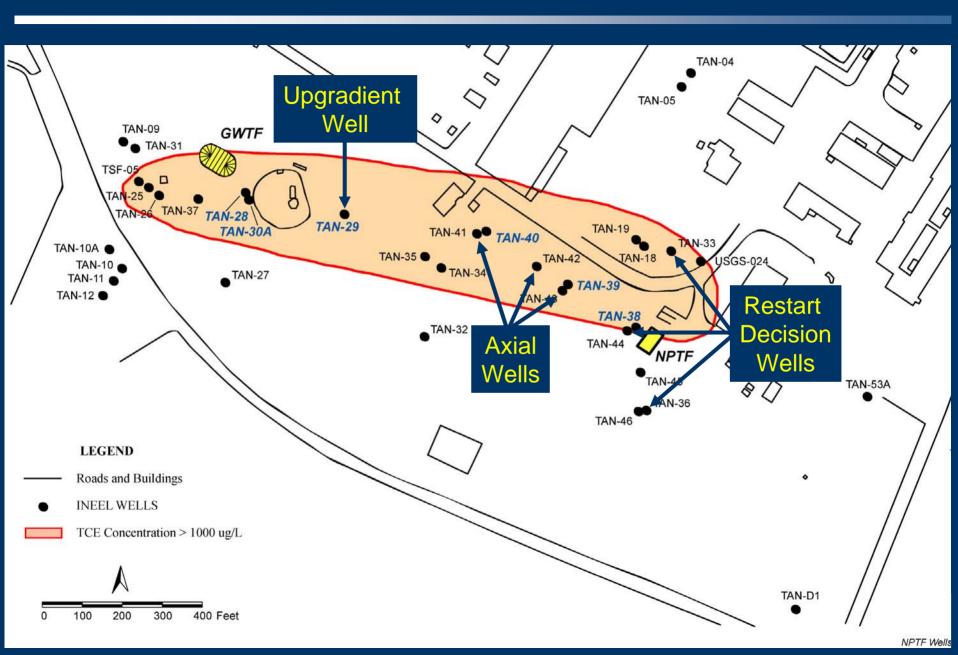


Accomplishments: Hotspot

- Increased the dissolution of source material available for biological degradation
- Increased the biomass of the biological population in and around the source area capable of biological degrading the contaminant
- Increased the area of influence, in this case, biological activity, surrounding (upgradient and down-gradient) of the residual source area

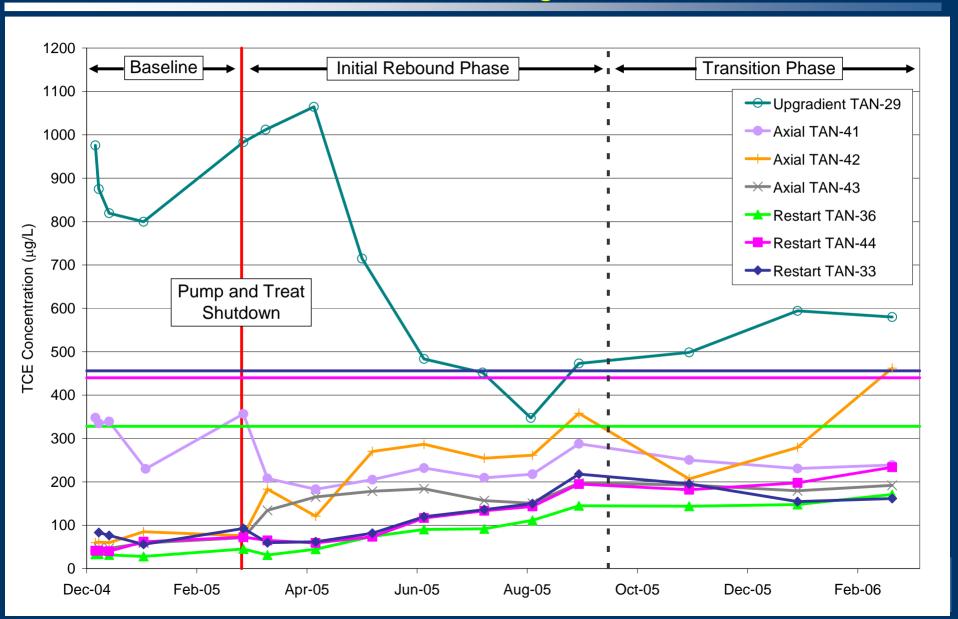


Medial Zone Rebound Test



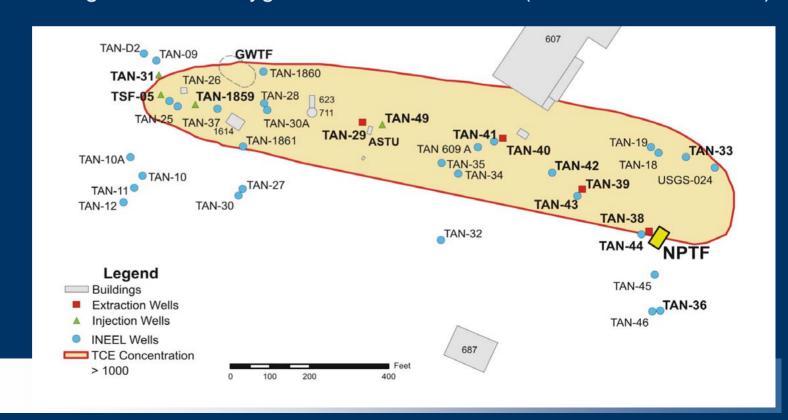
Rebound Test Results

12 months following shutdown



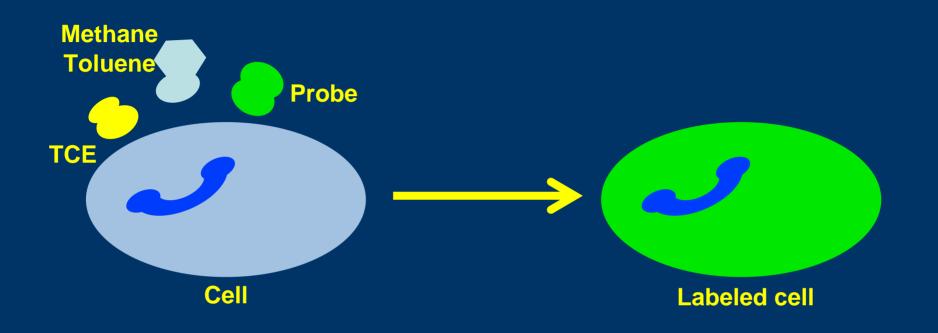
Activity Tools

- Quantitative PCR (qPCR): Target for this analysis is Dehalococcoides and respective degradative genes.
- Fluorescent in situ hybridization (FISH): Targets either 16S rDNA or RNA to determine the activity of the targeted organisms; wide array of target organisms.
- Enzyme Activity Probes (EAP): Detection of specific enzyme activities; probes validated target aromatic oxygenases and the sMMO (aerobic cometabolism).

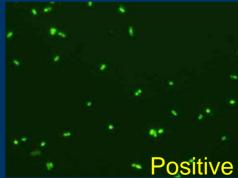




Enzyme Activity Probes



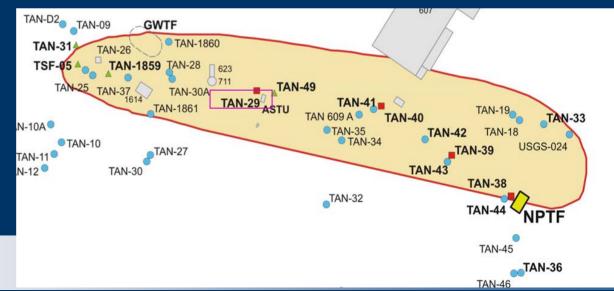






Enzyme Activity Probes in Medial Zone

	% Positive Probes					TCE	Methan	Dissolved Oxygen	
Well	DAPI	3НРА	PA	Cinn	Coumarin	(μ g/L)	μ g/L) e (μg/L)	(mg/L)	рН
TAN-28	6.07E+05	0.56	6.12	1.35	44	1061	7185	0.17	7.01
TAN-29	5.68E+05	4.75	7.9	0.72	57	364	5968	0.33	7.03
TAN-41	4.86E+05	0.69	10.21	3.44	63	259	1624	0.95	7.01
TAN-42	7.79E+05	1.03	6.15	1.07	35	365	320	8.13	7.04
TAN-43	1.30E+06	0.11	4.3	88.0	49	213	175	6.98	7.03
TAN-44	6.53E+05	6.66	12.33	0.43	55	234	110	6.91	7.15
TAN-33	4.03E+05	10.54	10.14	4.49	54	237	134	12.03	7.25
TAN-36	4.75E+05	6.88	7.05	0.89	63	175	122	9.32	7.18





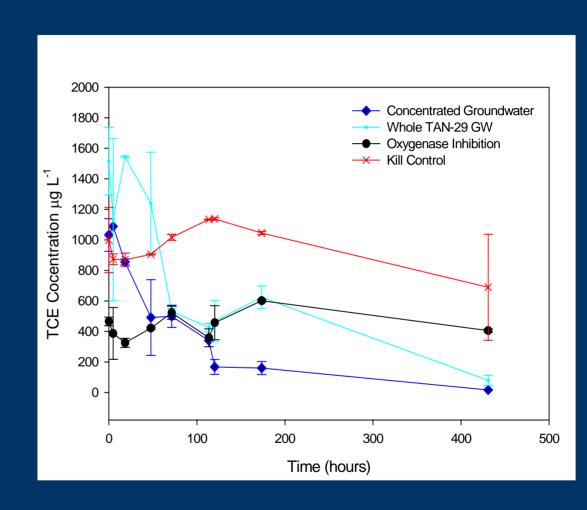
Aerobic Degradation Rate Evaluation

Evaluated rate of degradation for TCE at TAN-29

Determined activity of organisms with (a) aromatic oxygenases & (b) methane monooxygenase

Determined rate of removal for TCE over time

Control studies





Field Evaluation (*in situ*) of cometabolic degradation at TAN

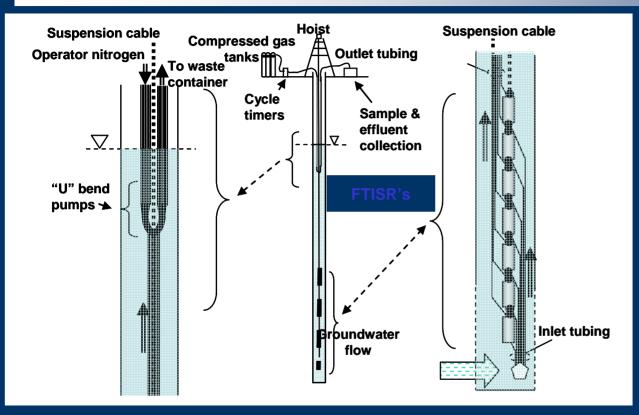


Diagram of FTISR reactors submerged in the groundwater. Each reactor is 6.35 cm (internal diameter) x 120 cm long with 3700 cm³ volume. Groundwater flow rate through each reactor is pneumatically controlled from the surface through "U" bends.

FTISR's were installed in November 2006 at TAN-35





Accomplishments: in situ evaluations

Proteomics:

- a new method (mass spectrometry) was developed for extracting total from environmental planktonic and/or biofilm samples
- detection of both sMMO and pMMO proteins in groundwater (TAN-29)

Real-time PCR:

- assays for detection of sMMO were developed
- studies indicate the presence of mmoX

RFLP (restriction fragment length polymorphism) analysis:

- planktonic and biofilm populations differ from one another



Accomplishments continued

Phylochip:

- high diversity for both groundwater and basalt biofilm populations
- detected methanogens and methanotrophs

PLFA (phospholipid fatty acid):

- signatures for both Type I and Type II methanotrophs

Enzyme probes:

- sMMO enzyme activity across the medial zone
- results from microcosm study suggest these processes may contribute to the removal of TCE from the medial zone at TAN.

Together these studies provide multiple lines of evidence supporting the presence and activity of methanotrophic populations in the SRPA; which provides additional evidence that NA is occurring in groundwater at the TAN site.



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